Topic B – Part 2 Proteins (3Hrs)

IB Chemistry

Topic B – Biochem

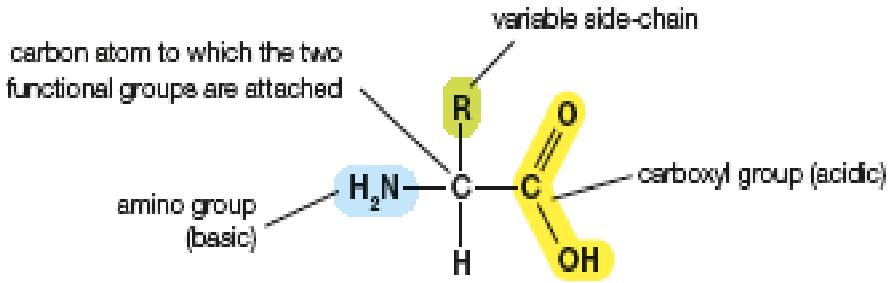
B2 Proteins - 3 hours

- B.2.1 Draw the general formula of 2-amino acids. (1)
- B.2.2 Describe the characteristic properties of 2-amino acids (2)
- B.2.3 Describe the condensation reaction of 2-amino acids to form polypeptides. (2)
- B.2.4 Describe and explain the primary, secondary ('-helix and (-pleated sheets), tertiary and quaternary structure of proteins. (3)
- B.2.5 Explain how proteins can be analyzed by chromatography and electrophoresis. (3)
- B.2.6 List the major functions of proteins in the body. (1)

B2.6 - Proteins

Proteins

- Proteins are macromolecules that are **polymers** of amino acids. They have two main roles
- **Structural Proteins**; proteins go into making muscle tissue, connective tissue, and skin, hair, and nails, just to name a few.
- **Function Proteins;** proteins are enzymes which catalyze biochemical reactions
  - Building up macromolecules requires energy and an enzyme lowers the amount of energy that is necessary.
  - Also as messengers known as hormones



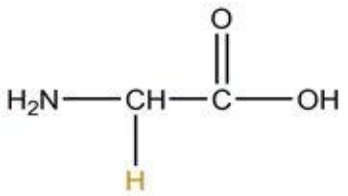
B2.6 – Proteins Function

Role of Protein	Example	Specific Function
Structural	Keratin	Protective covering in hair and fingernails
Structural	Collagen	Connective tissue in skin and tendons
Structural	Myosin	Contractile action in muscles to bring about movement
Enzyme (catalyst)	Lactase	Hydrolyses lactose into glucose and glactose
Hormone	Insulin	Controls and maintains the concentration of glucose in the blood
Protective mechanisms	immunoproteins	Act as antibodies which help destroy foreign proteins (e.g. from bacteria) in the blood
Transport molecules	Hemoglobin	Carries oxygen from the lungs to all respiring cells
Storage molecules	Casein	Food substance in milk
Lubrication	Mucoproteins	Mucous secretions to reduce friction in many parts of the body, e.g. the knee joint

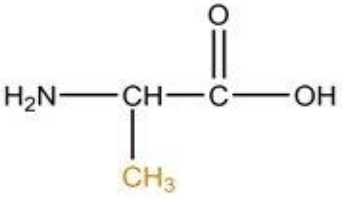
B2.1 - Amino Acids and Proteins

- Amino Acids are the building blocks of proteins
- There are 20 amino acids that go into producing proteins.
  - Each have a carboxylic acid AND an amino group
    - COOH, loses an OH group and NH_2 loses an H, forming a bond between two amino acids and water.
  - These amino acids are polymerized by a dehydration synthesis to form long chains of repeating amino acids called a protein. (taking water out)
  - The arrangement of the amino acids in the polymer determine the structure of the protein which confers to it is function or structural attributes.
    - Meaning the shape/order/structure of the amino acids in a protein determines it's function

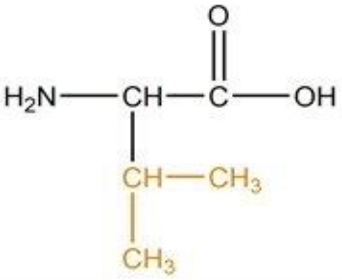
Gly



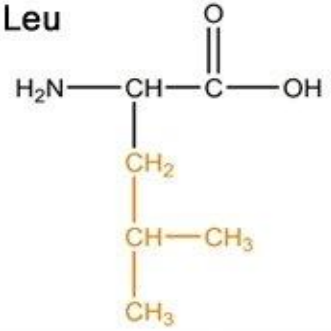
Ala



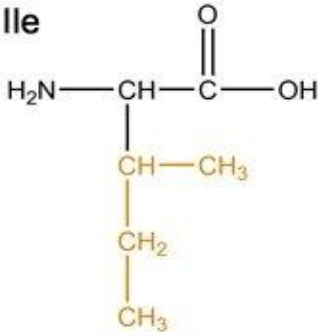
Val



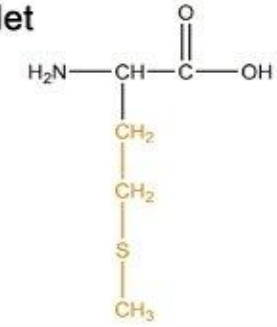
Leu



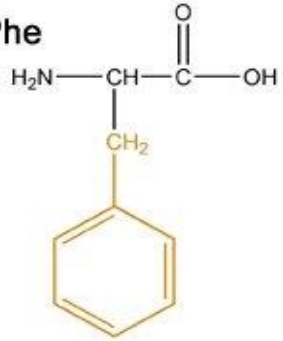
Ile



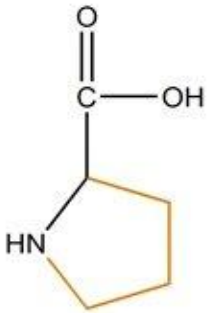
Met



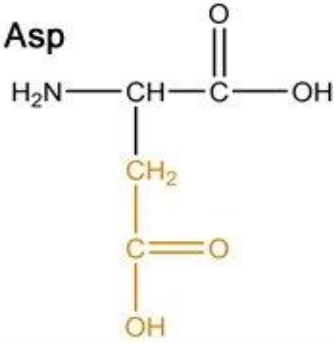
Phe



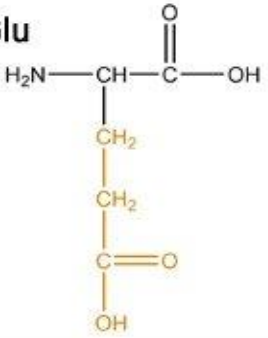
Pro



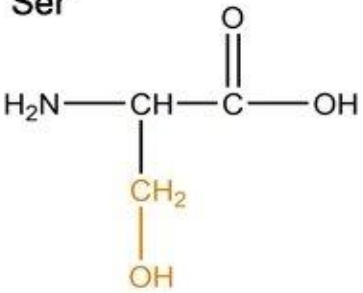
Asp



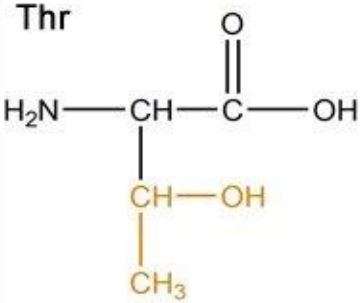
Glu



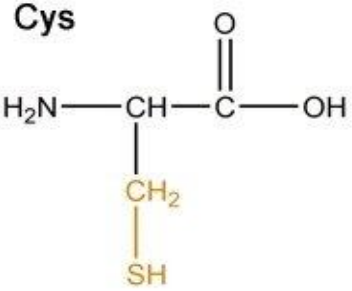
Ser



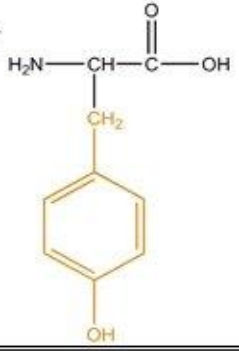
Thr



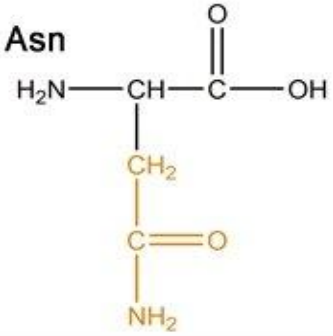
Cys



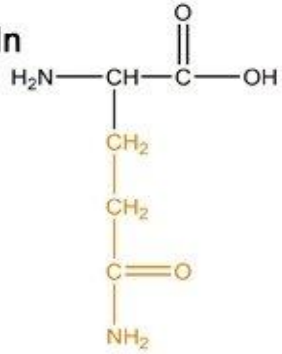
Tyr



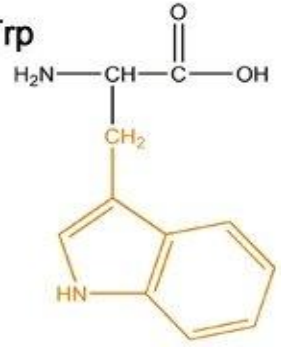
Asn



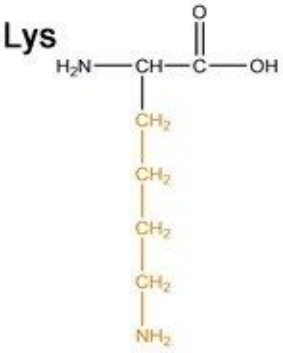
Gln



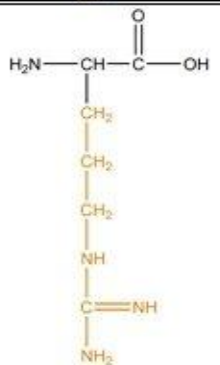
Trp



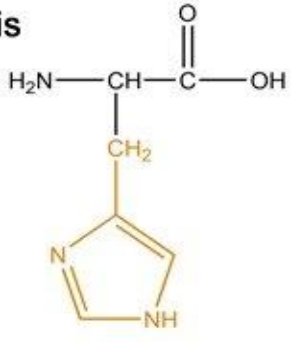
Lys



Arg



His



B2.1 - Essential Amino Acids

- Of the 20 amino acids that make up proteins 10 of them can be synthesized by the human body
- The other 10 amino acids must be acquired from food sources. These amino acids are known as essential amino acids

B2.1 - Essential Amino Acids

Essential amino acids

- Arginine
- Histidine
- Isoleucine
- Leucine
- Lysine
- Methionine
- Phenylalanine
- Threonine
- Tryptophan
- Valine

Non-Essential amino acids

- Alanine (from pyruvic acid)
- Asparagine (from aspartic acid)
- Aspartic Acid (from oxaloacetic acid)
- Cysteine
- Glutamic Acid (from oxoglutaric acid)
- Glutamine (from glutamic acid)
- Glycine (from serine and threonine)
- Proline (from glutamic acid)
- Serine (from glucose)
- Tyrosine (from phenylalanine)

B2.1 - Essential Amino Acids

Complete protein

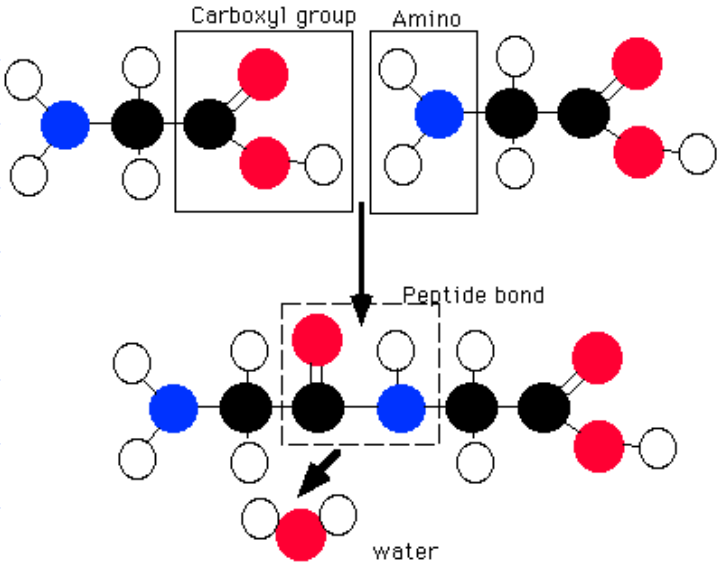
- Contains all 10 essential amino acids
- Proteins derived from animal sources are complete proteins
- Beans contain some complete protein as well

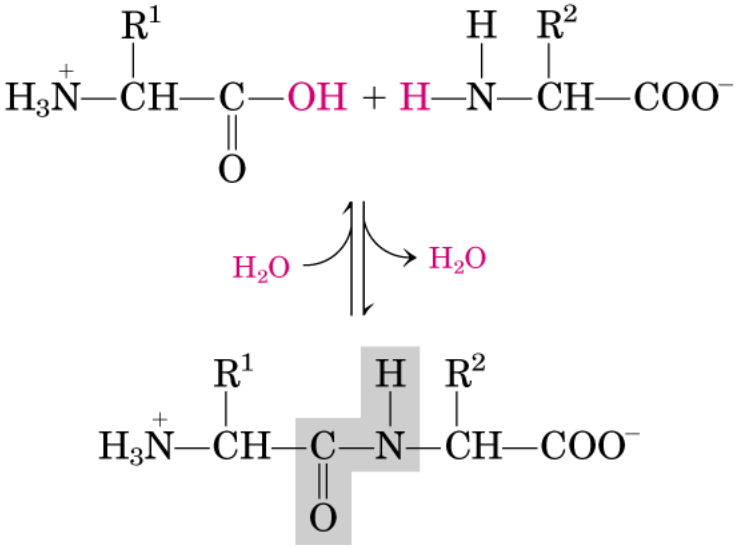
Incomplete protein

- Lack one of more of the essential amino acids
- Most vegetable proteins are incomplete proteins
- Beans are an exception to this generalizations

B2.3 – Condensation, dehydration synthesis

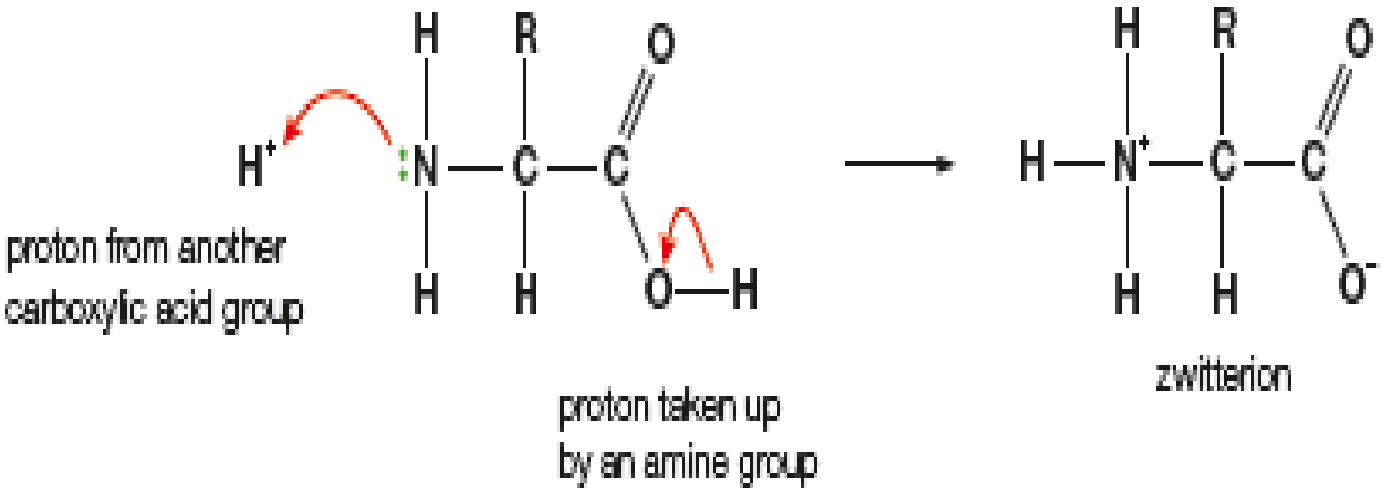
- With three-letter abbreviations.
- These twenty amino acids that make up proteins, can be linked together through dehydration synthesis.
- The carboxyl group of one amino acid bonds with the amino group of a second acid to yield a **dipeptide** and water. Proteins are long chains of amino acids linked by peptide bonds.





B2.2 - Amino Acids are Amphoteric

- Amino acids are **amphoteric.** They are capable of behaving as both an acid and a base, since they have both a proton donor group and a proton acceptor group.

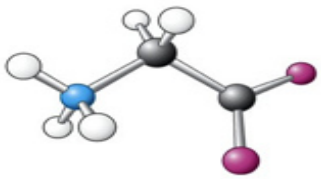


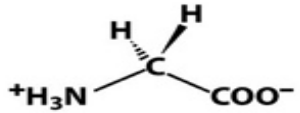
- In neutral aqueous solutions the proton typically migrates from the carboxyl group to the amino group, leaving an ion with both a (+) and a (-) charge.

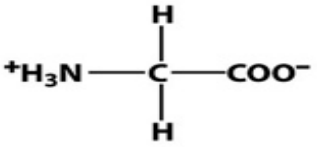
B2.2 – (1) The Zwitterion

- Crystaline amino acids have relatively high melting or decomposition points.
- Soluble in polar solvents.
- Amino acids exists as dipolar ions known as **Zwitterions.**
- Internal acid-base reaction, a hydrogen ion is transferred fro the carboxylic acid to the amino group.

Glycine (Gly, G)

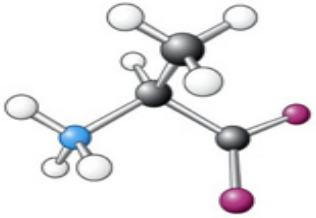


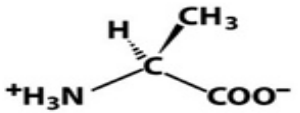


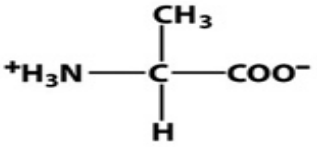


Glycine (Gly, G)

Alanine (Ala, A)





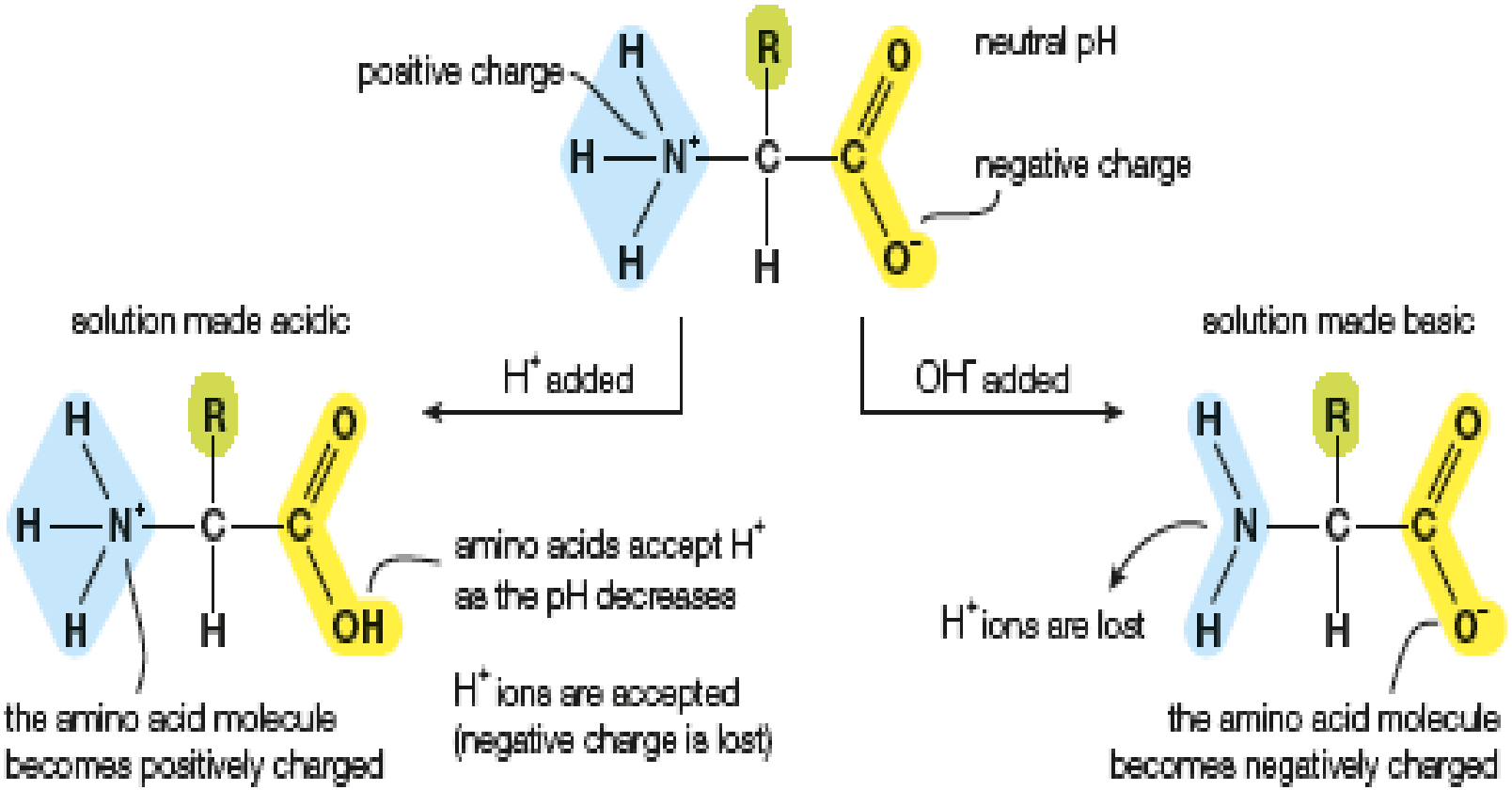


Alanine (Ala, A)

B2.2 – (2) Buffer Action

- In aqueous solution the amino and carboxylic acid functional groups both ionize or dissociate
- Carboxylic group releases hydrogen ions and hence acts as a Bonsted-Lowry acid
  - $-COOH\ (aq) \leftrightarrow -COO^{-}\ (aq) + H^{+}\ (aq)$
- The amino group can accept a hydrogen ion from solution and so acts as a Bronsted-Lowry base
  - $-NH_2\ (aq) + H^{+}\ (aq) \leftrightarrow -NH_3^{+}\ (aq)$
- In solutions of neutral (both dissociated), acidic (low pH, amino acid accepts hydrogen, +), basic (high pH, amino acid donates hydrogen, -)
- Consequently, amino acids regulate the pH of a system by "mopping up or donating H^{+} ions)

B2.2 – (2) Buffer Action 2

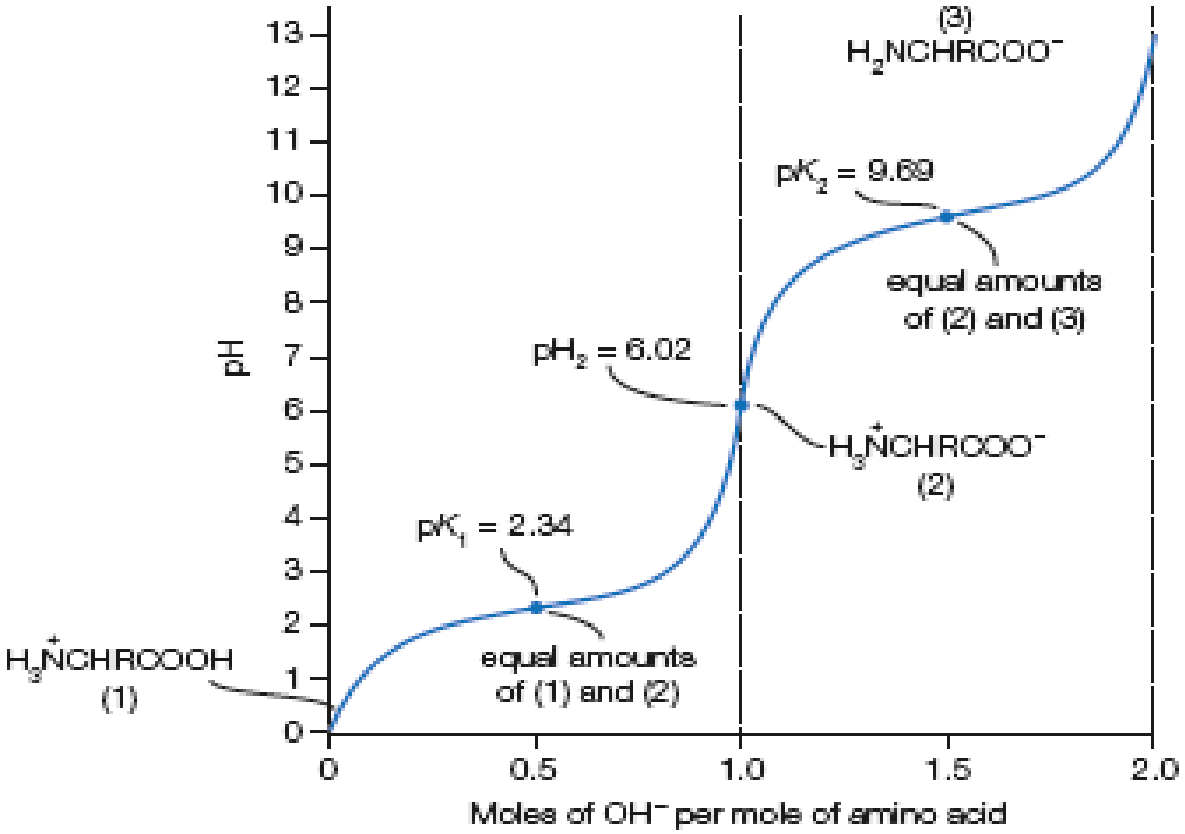


B2.2 – (3) Isoelectric Point

- Aminio acids such as alanine (where R is a methyl group) are dibasic when it's fully deprotonated. It can donate two protons H^+ when in titration with a strong base.
  - $H_3N^+CH(CH_3)COOH(aq) + OH^-(aq) \rightarrow H_3N^+CH(CH_3)COO^-(aq) + H_2O(aq)$
  - $H_3N^+CH(CH_3)COO^-(aq) + OH^-(aq) \rightarrow H_2NCH(CH_3)COO^-(aq) + H_2O(aq)$
- The isoelectric effect is the basis for a separation technique known as **electrophoresis**.

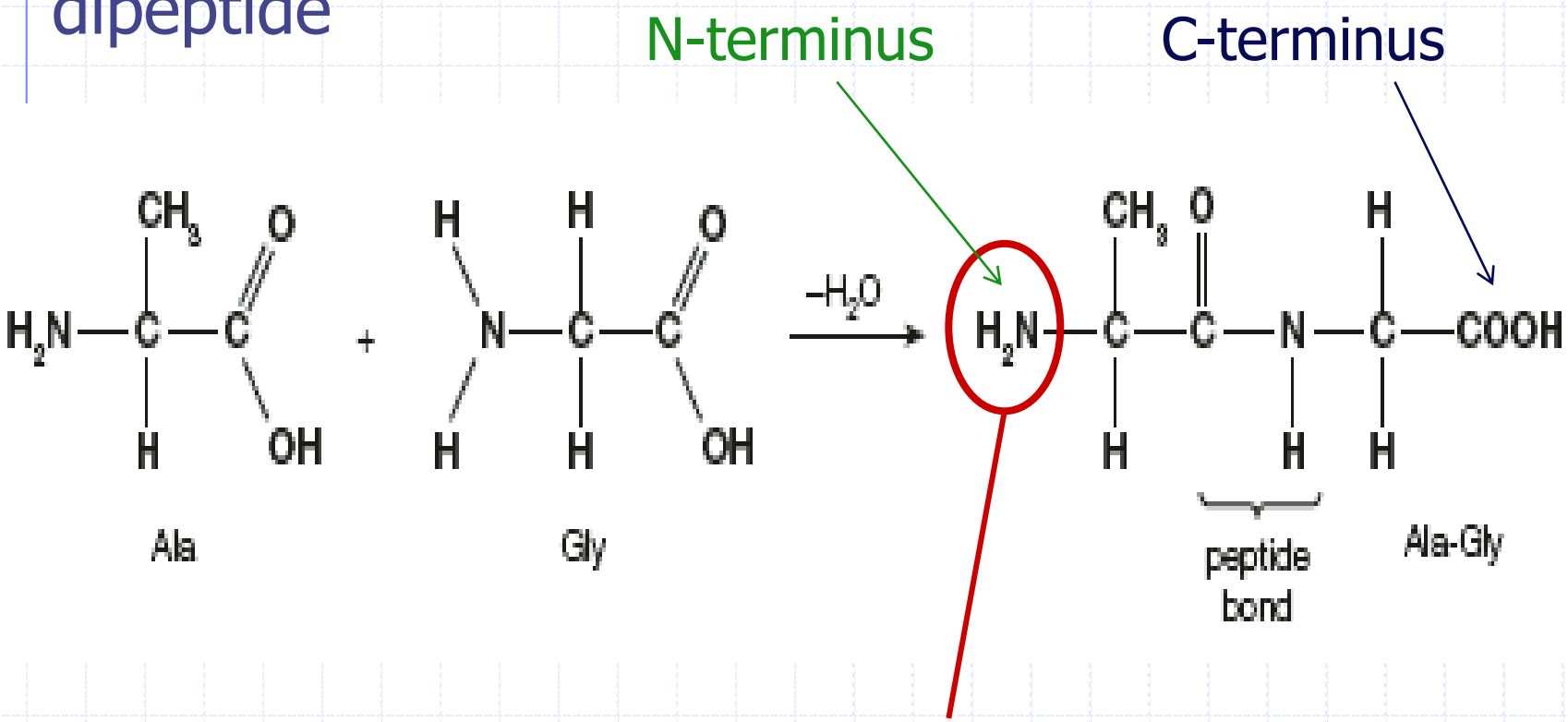
B2.2 – (3) Isoelectric Point 2

- The first part of the curve is the first reaction, the second, the second reaction. The flat parts of the curve correspond to the buffering regions where pH does not vary significantly.



B2.3 – Forming Polypeptides

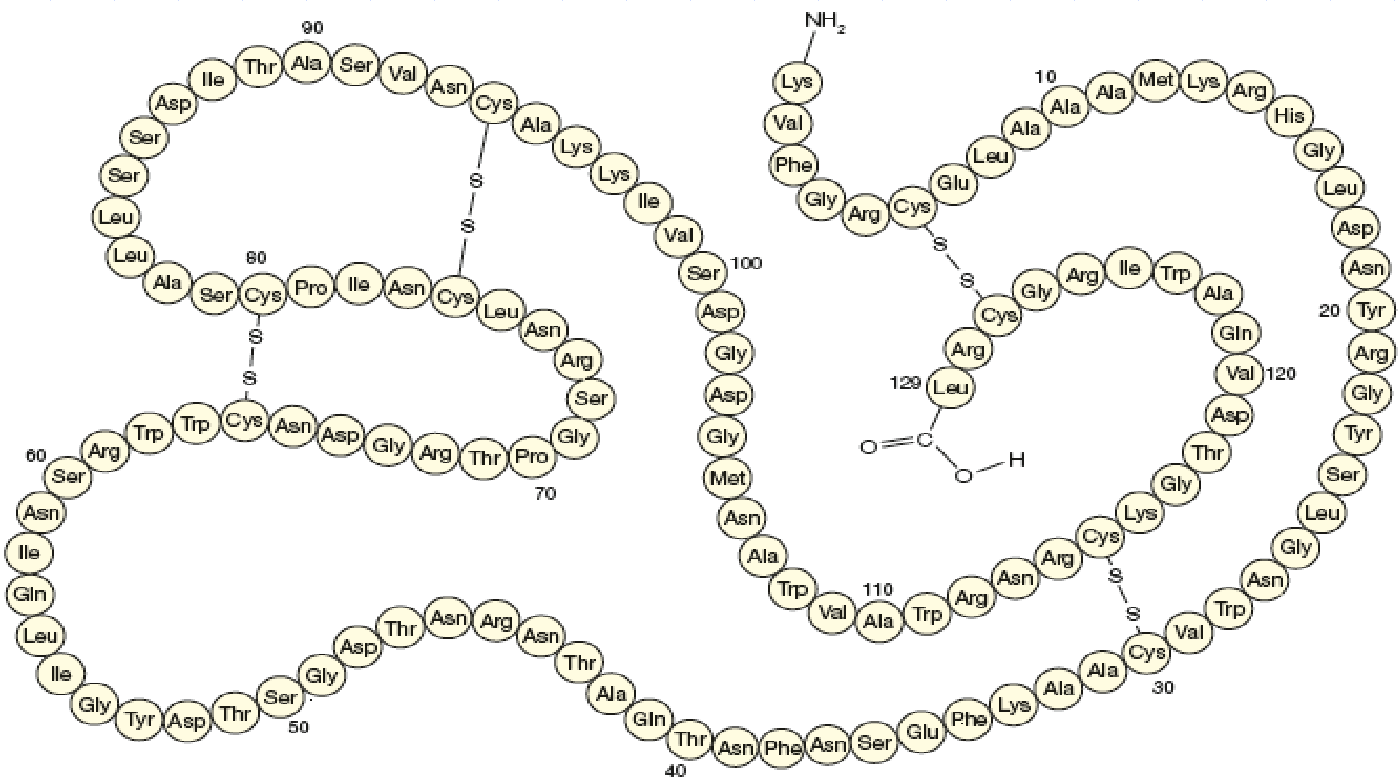
- Two amino acids can combined to form a simple dipeptide



By convention, when drawing polypeptide chains the amino group is written on the left

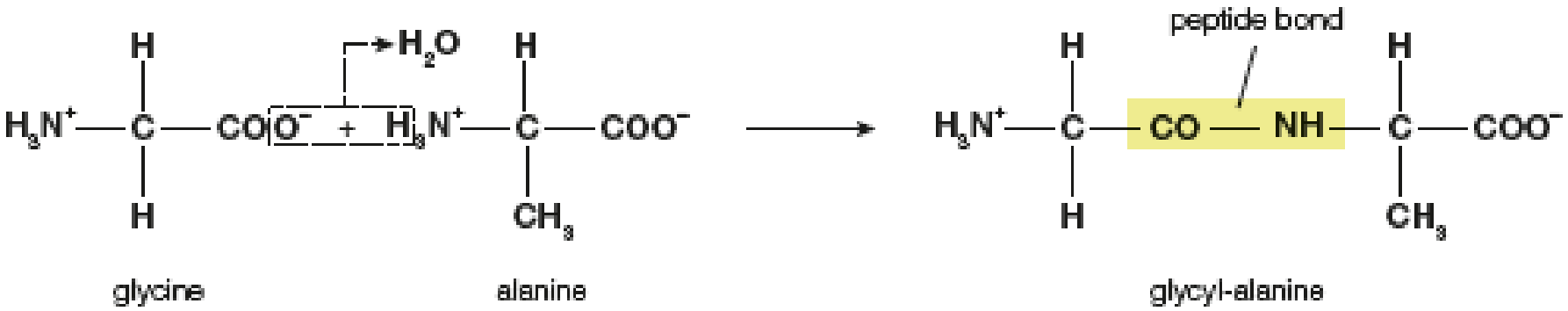
B2.3 – Forming Polypeptides

- Chains of amino acids can form polypeptides (proteins) that are 1000's of AA's long
- We will get into the folding and structure of proteins in B2.4 on Friday, please read ahead.

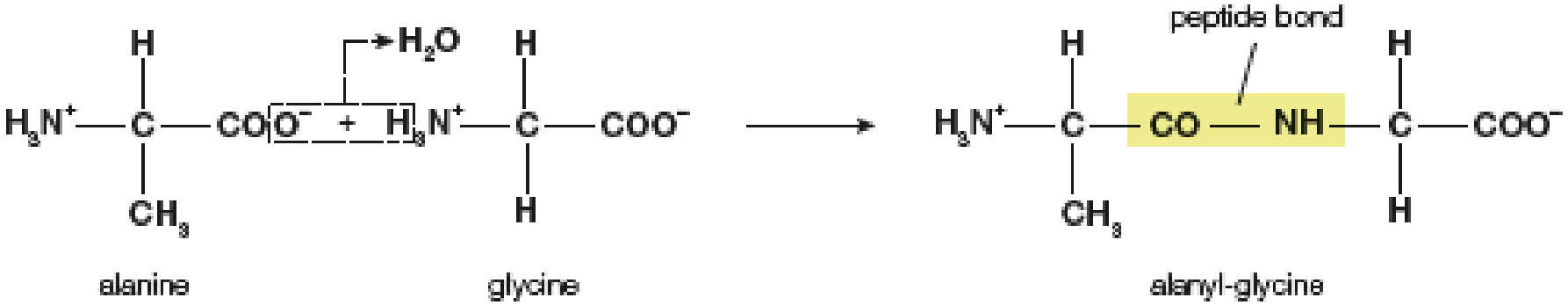


B2.3 - Gly-Ala OR Ala-Gly

For example, glycine and alanine can react like this:



But if the amino group of glycine reacts with the carboxyl group of alanine, a different dipeptide, alanyl-glycine, is formed



B2.3 – **Amino Acid Residue**

- A peptide (protein) is NOT made up of amino acids, rather it is made up of amino acid leftovers (**residue**).
- When amino acids form peptide bonds together they give off water, losing H or OH each. They are no longer the same structure
- For this reason we use the term **amino acid residue**

B2.3 – Reversible?

- The process of forming a peptide bond can be reversed with the use of a strong acid in the presence of water.

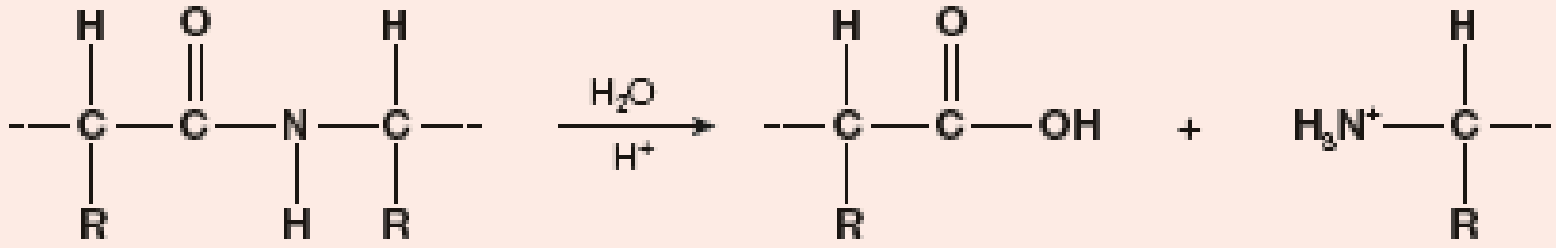
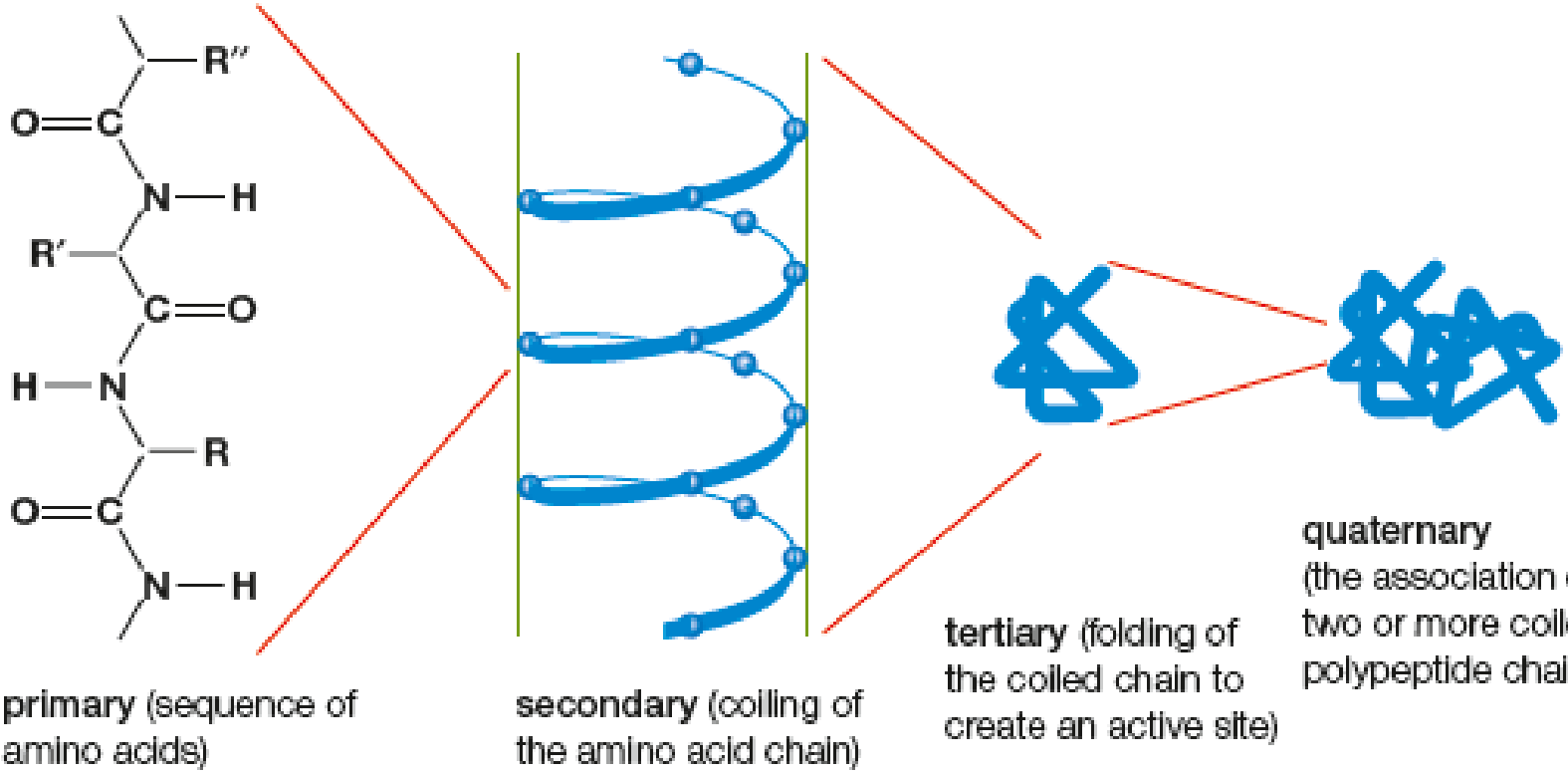


Figure 22.14 Acid hydrolysis of proteins

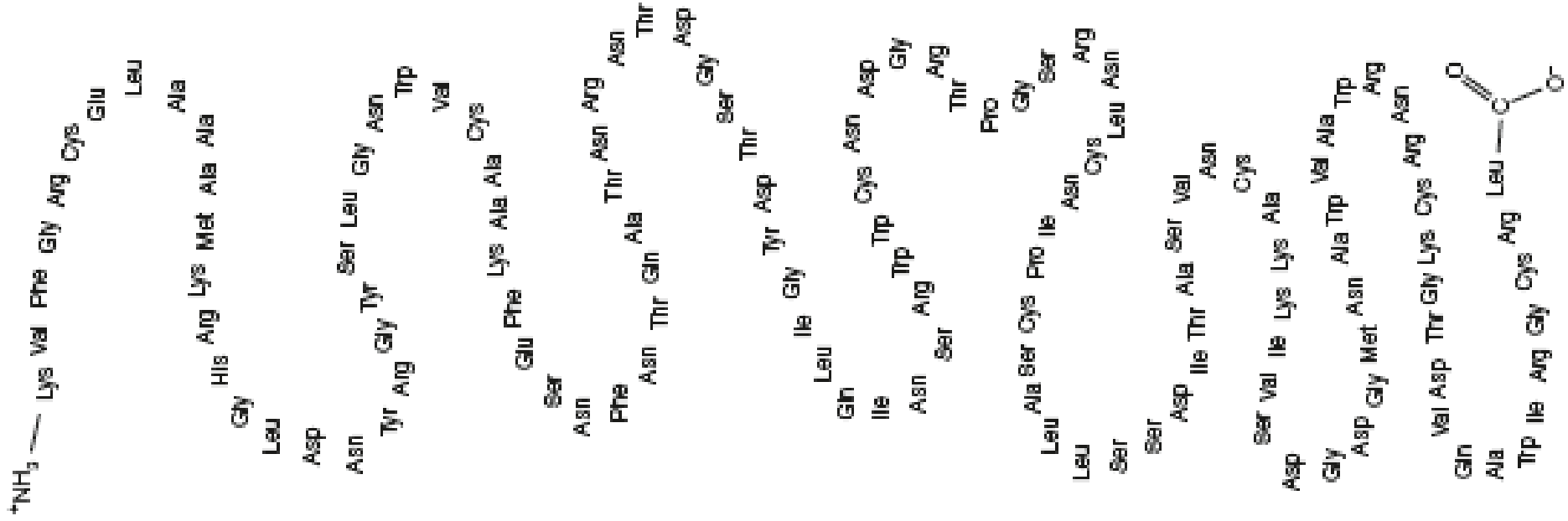
B2.4 – Protein Structure

- There are four main contributing factors to the structure of proteins, primary, secondary, tertiary, and quaternary



B2.4 – **Primary Structure**

- The linear sequence of amino acids in the polypeptide chain
- Change in a single amino acid (such as a mutation) can cause drastic differences in function

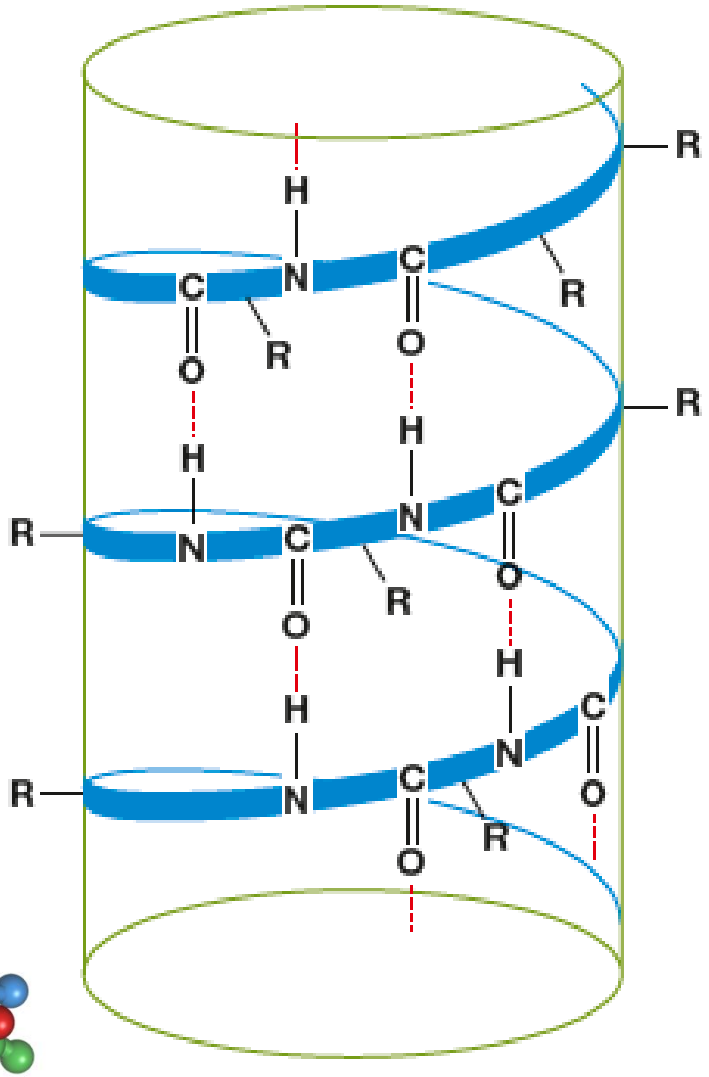


B2.4 – **Secondary Structure**

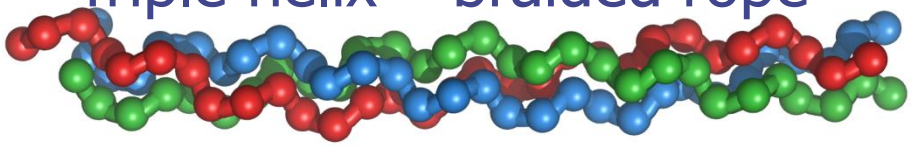
- Secondary, refers to the regular and permanent arrangement of sections of the polypeptide chain, both are **stabilized by hydrogen bonds**
  - α-helix (alpha, spiral)
  - β-sheet (beta, sheets)

B2.4 – Secondary/α-helix

- The coiled conformation of the polypeptide chain.
- Right-handed
- -R groups on outside and are perpendicular
- N (in peptide bond) bonded to O (of peptide carbonyl group, four residues down the chain.

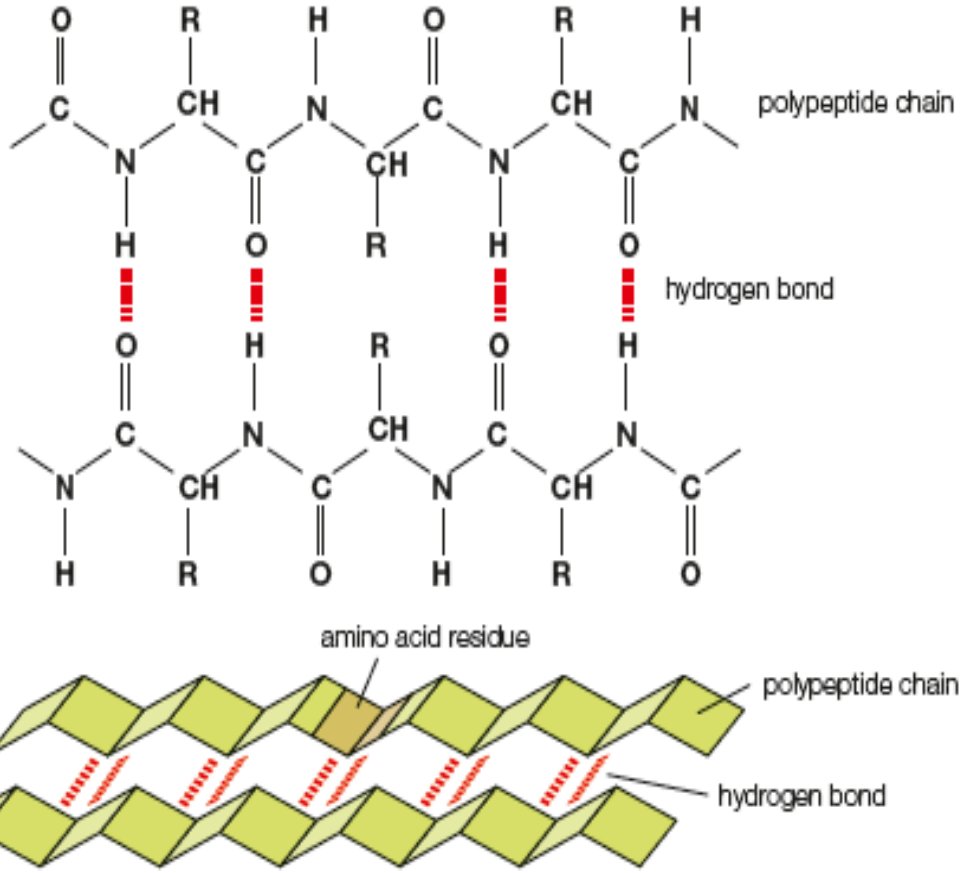


Triple helix – braided rope



B2.4 – Secondary/β-sheet

- Formed by polypeptides whose amino acid residues have small and compact side-chains.
- Composed of adjacent polypeptide chains (of the same protein) side-by-side and connected with hydrogen bonding



Learning Check 1

Indicate the type of structure as

(1) primary
(2) alpha helix
(3) beta pleated sheet
(4) triple helix

A. Polypeptide chain held side by side by H bonds
B. Sequence of amino acids in a polypeptide chain
C. Corkscrew shape with H bonds between amino acids
D. Three peptide chains woven like a rope

Solution 1

Indicate the type of structure as

(1) primary (2) alpha helix

(3) beta pleated sheet (4) triple helix

A. 3 Polypeptide chain held side by side by H bonds

B. 1 Sequence of amino acids in a polypeptide chain

C. 2 Corkscrew shape with H bonds between amino acids

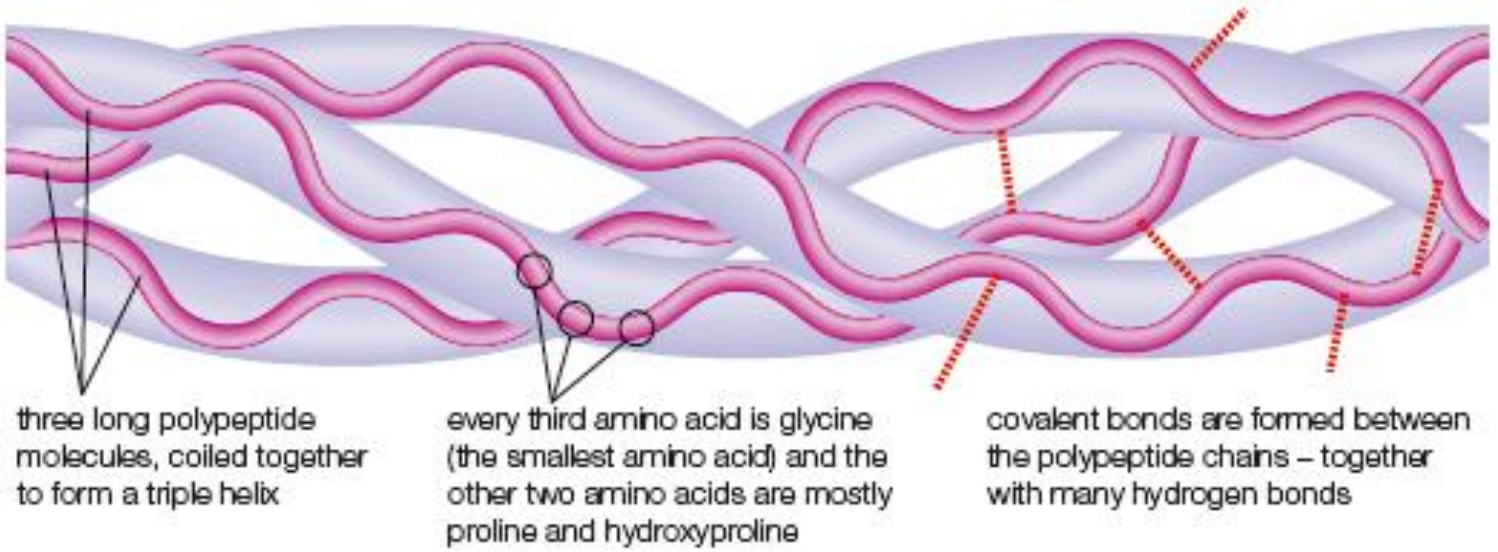
D. 4 Three peptide chains woven like a rope

B2.4 – Tertiary Structure

- Overall 3D shape of a single protein.
- Held by a specific shape by hydrogen bonds and van der Waala (intermolecular) and disulfide bonds, ionic bonds (intramolecular forces) involving side-chains.
- Proteins can be classified as
  - Fibrous: long molecules arranged to form fibers
  - Globular: Folded into a compact and precise shape

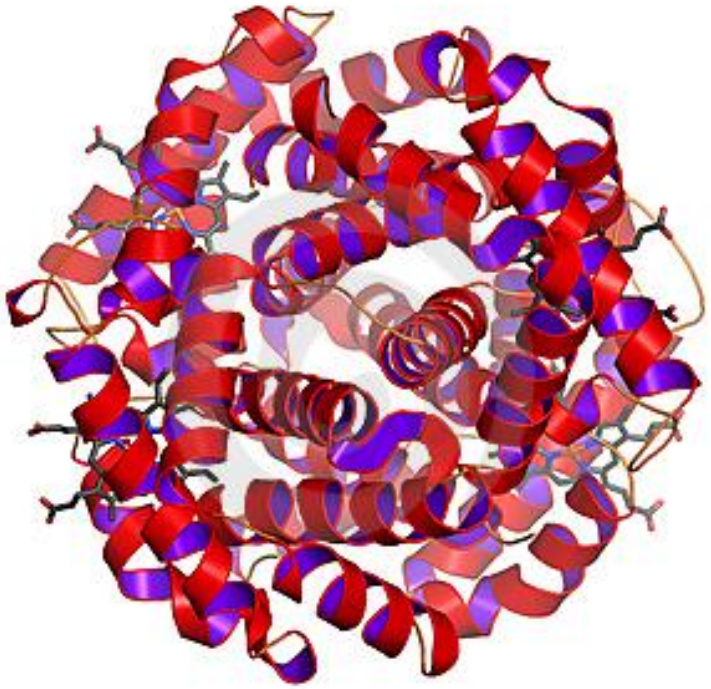
B2.4 – Tertiary / Fibrous

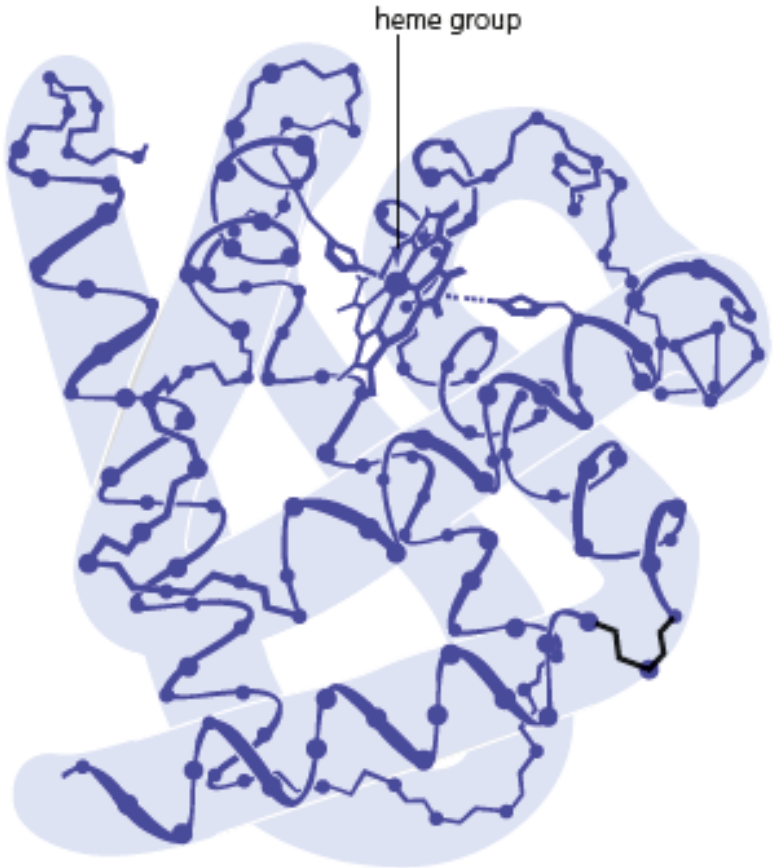
- Fibrous: long molecules arranged to form fibers
  - Collogen is the most abundant fibrous protein in the human body



B2.4 – Tertiary / Globular

- Globular: Folded into a compact and precise shape, soluble in water generally





myoglobin

Globular and Fibrous Proteins

Globular proteins

"spherical" shape

- **Insulin**
- **Hemoglobin**
- **Enzymes**
- **Antibodies**

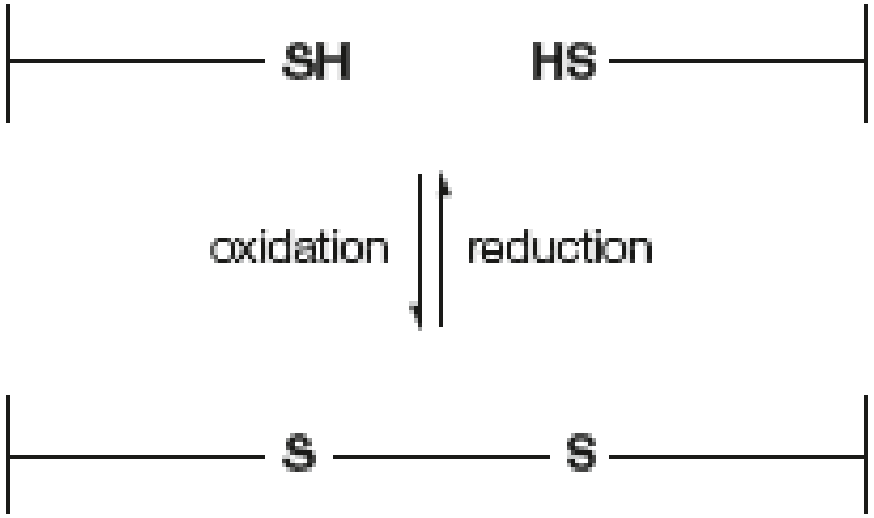
Fibrous proteins

long, thin fibers

- **Hair**
- **Wool**
- **Skin**
- **Nails**

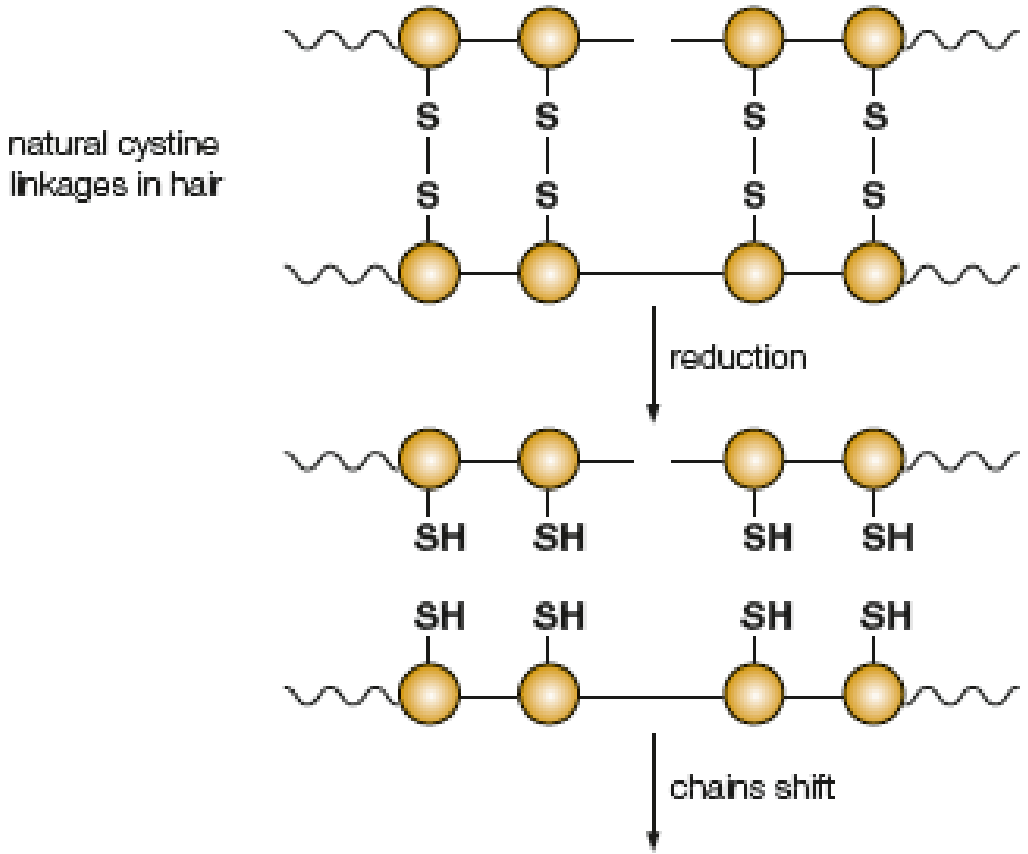
B2.4 – Tertiary/Di-sulfide Bridge

- This also includes any covalent cross links
  - Disulfide bridges –S–S– can form between two cysteine amino acid side chains.



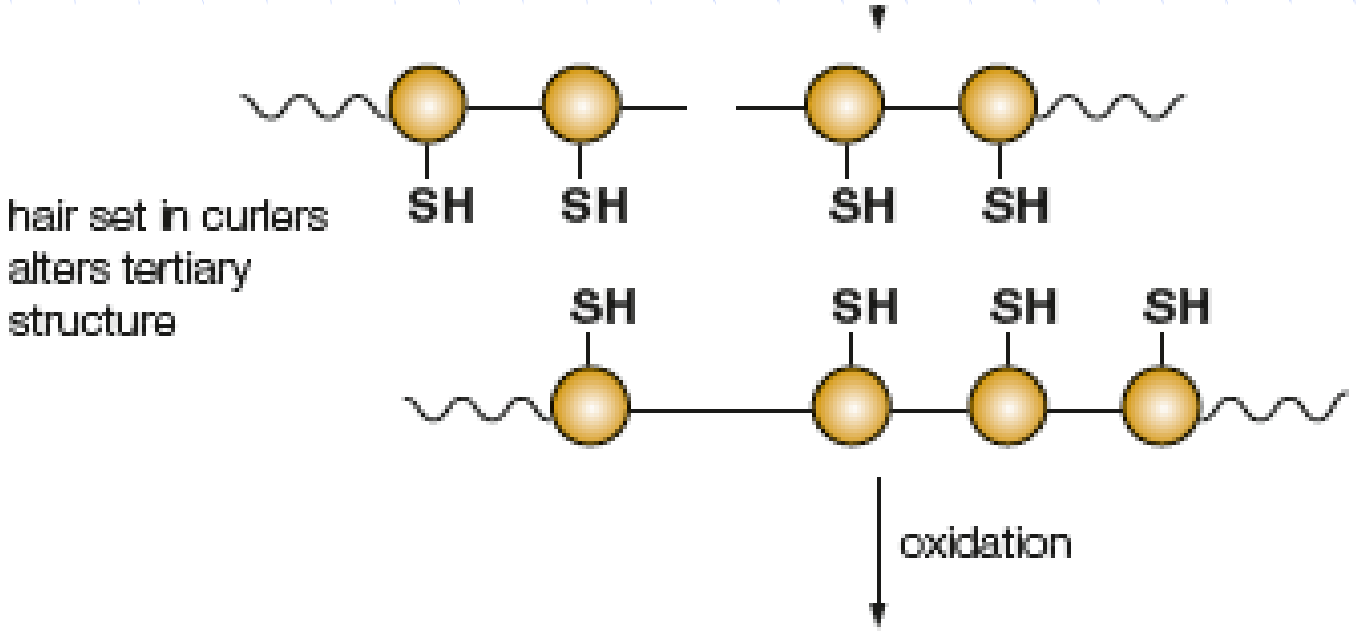
B2.4 – Tertiary/Human Hair(1)

- The curled or straight nature of hair is due to disulfide bonds.
- Hair is made of keratin, which includes the amino acid cystine, which has a thiol (-SH group).



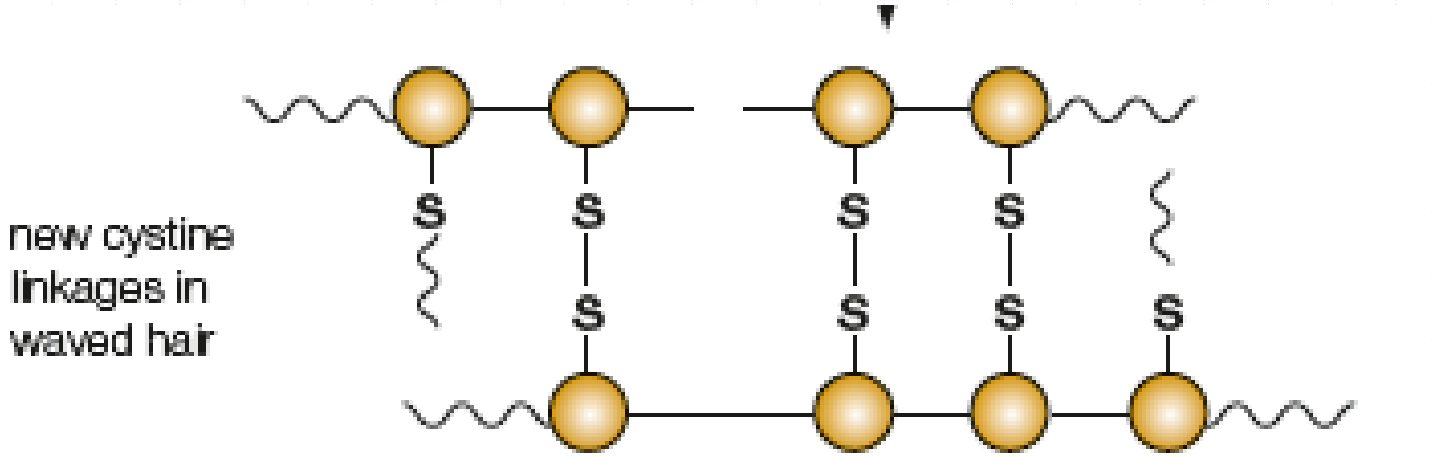
B2.4 – Tertiary/Human Hair(2)

- Giving hair a perm involves breaking and re-forming of the disulfide bonds in a two stage process:
  1. Hair is treated with solution of thiol to disturb –SH bonds. Hair is then set into required style, usually with rollers.



B2.4 – Tertiary/Human Hair(3)

2. Then a formation of new disulfide bonds by oxidation of the thiol groups which set the new style "permanently" into position.



B2.4 – Tertiary Structure

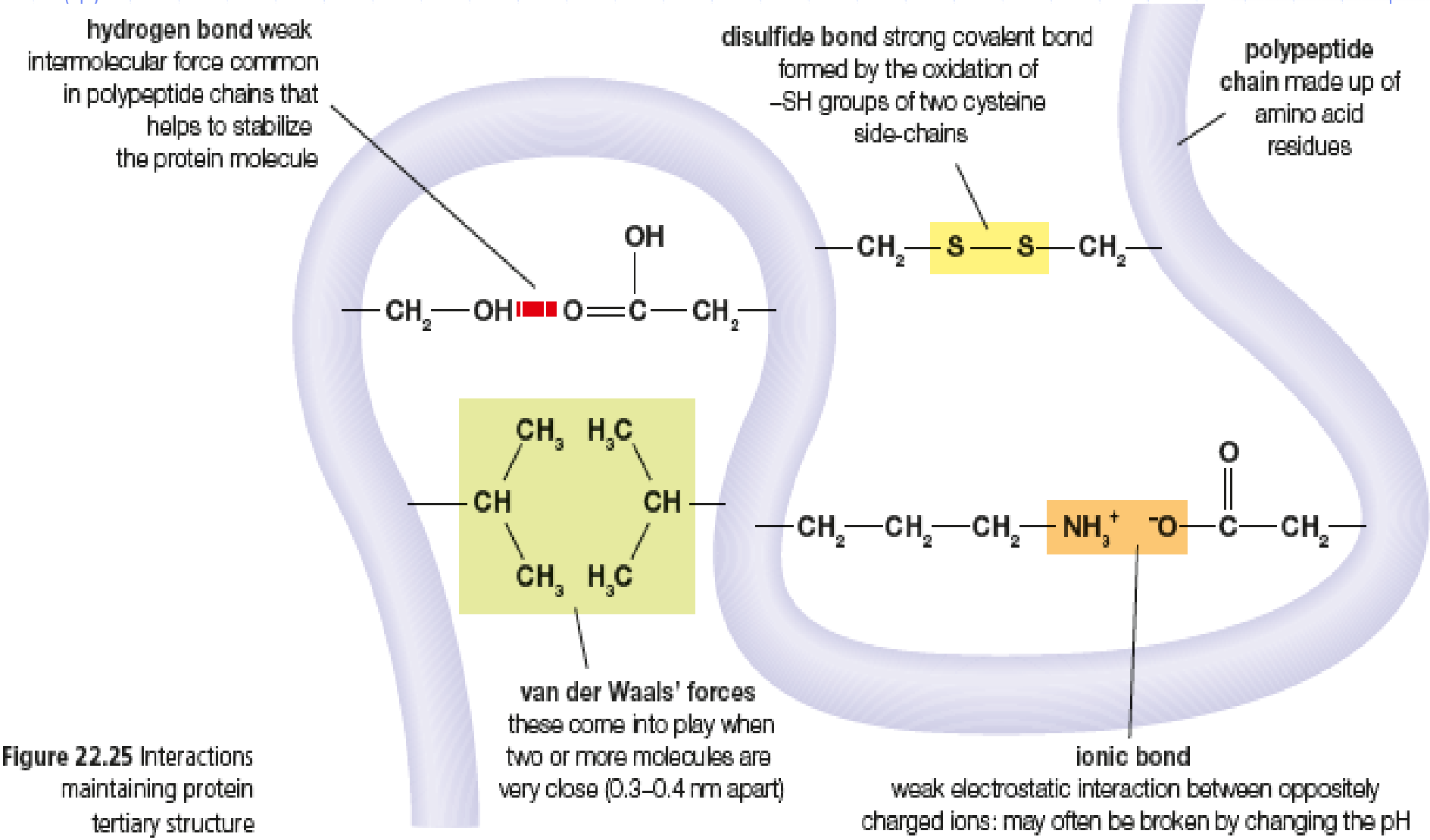


Figure 22.25 Interactions maintaining protein tertiary structure

Learning Check 2

Select the type of tertiary interaction as

(1) disulfide
(2) ionic
(3) H bonds
(4) hydrophobic

A. Leucine and valine
B. Two cysteines
C. Aspartic acid and lysine
D. Serine and threonine

Solution 2

Select the type of tertiary interaction as

(1) disulfide (2) ionic

(3) H bonds (4) hydrophobic

A. 4 Leucine and valine

B. 1 Two cysteines

C. 2 Aspartic acid and lysine

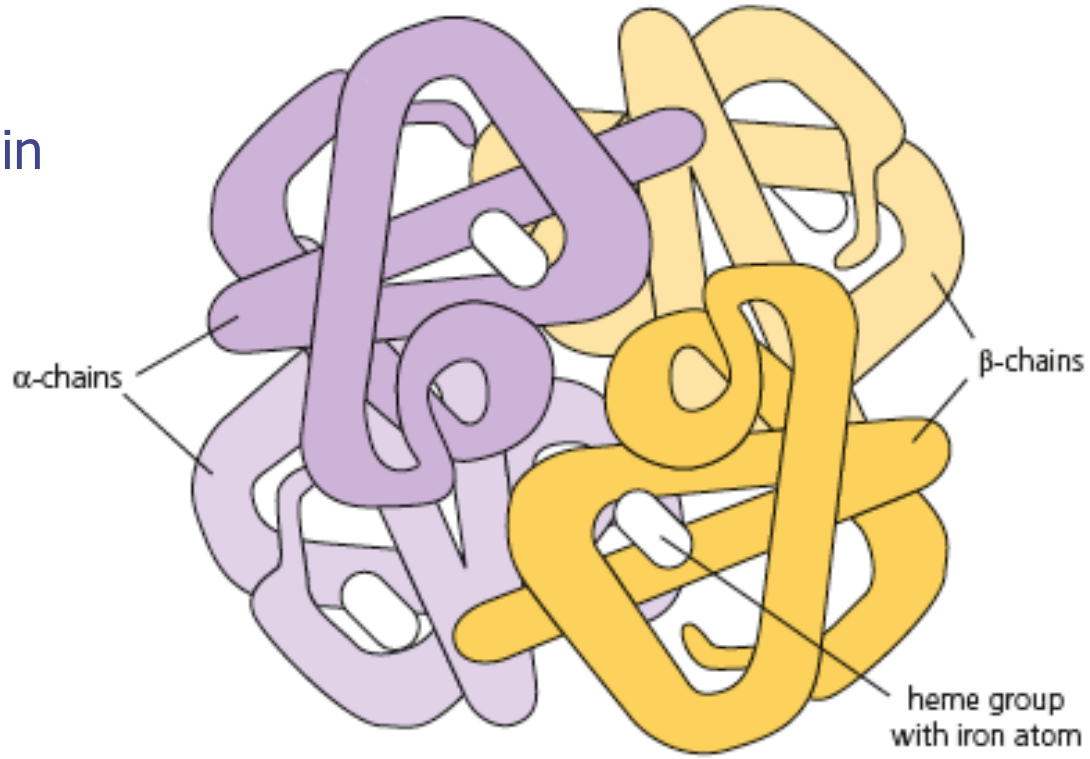
D. 3 Serine and threonine

B2.4 – **Quaternary Structure**

- When a protein consists of two or more tightly bound polypeptide chains, the overall conformation or shape is referred to as the **quaternary structure.**

Structure of Hemoglobin

- 2 α-chains
- 2 β-chains



Learning Check 3

Identify the level of protein structure

1. Primary
2. Secondary
3. Tertiary
4. Quaternary

A. Beta pleated sheet
B. Order of amino acids in a protein
C. A protein with two or more peptide chains
D. The shape of a globular protein
E. Disulfide bonds between R groups

Solution 3

Identify the level of protein structure

1. Primary
2. Secondary
3. Tertiary
4. Quaternary

A. 2 Beta pleated sheet
B. 1 Order of amino acids in a protein
C. 4 A protein with two or more peptide chains
D. 3 The shape of a globular protein
E. 3 Disulfide bonds between R groups

B2.4 - Denaturation

- Enzymes (proteins) function within a narrow range of pH and temperature values.
- High or low values (pH or T) cause the protein to go through a physical change **denature.**
- This involves loss of tertiary and quaternary structures, and is although often reversible since the primary and secondary structures remain the same, will change the function of the protein during this time.

B2.4 – Protein Folding Disorders

- The structure of a protein and it's ability to carry out it's biological function are so strongly correlated that even small structural defects can lead to a number of protein folding disorders.
  - Sickle cell anemia (single residue mutation)
  - Bovine spongiform encephalopathy (BSE) or "mad cow disease."
    - Disorder leads to the buildup of insoluble protein in the brain

B2.4 – Denaturing / Flip

- Creutzfeld-Jacob disease (CJD) is the human equivilent of "mad cow disease".
- Researchers have found that normal prion protins in the brain consist of α-helicies, but in CJD these proteins 'flip' and unfold into a protein with β-sheets, now insoluble, collect in the brain

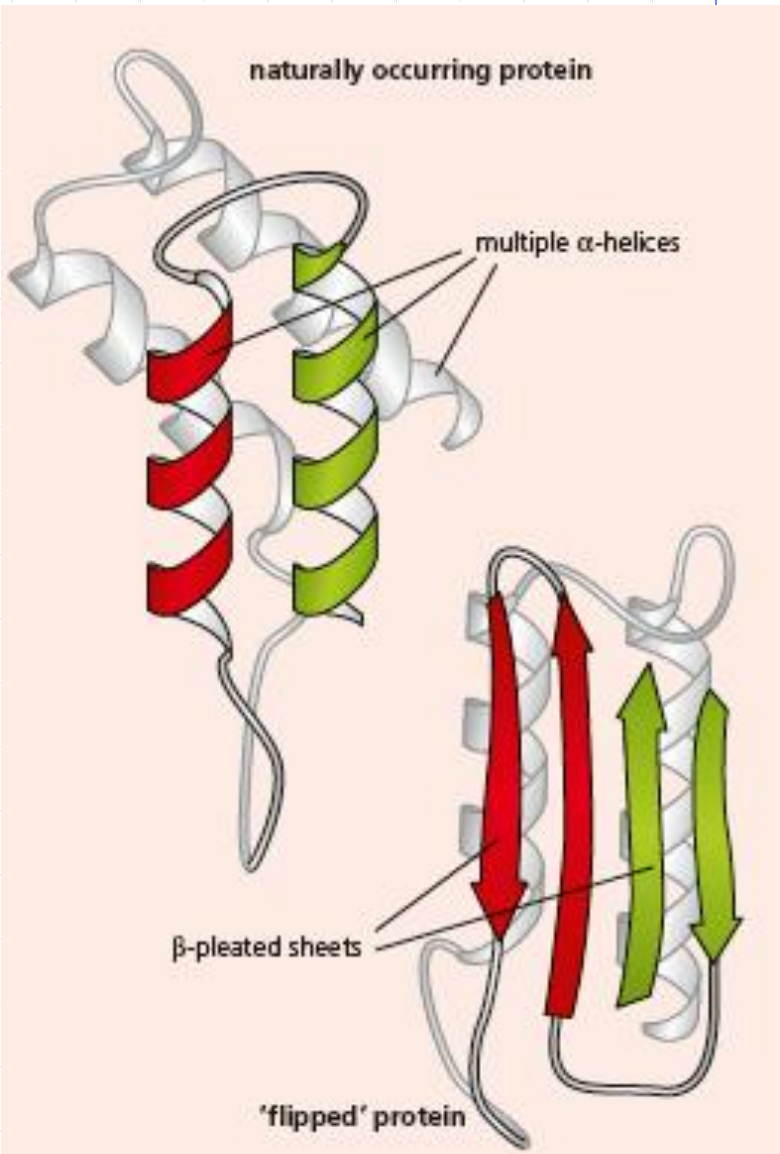
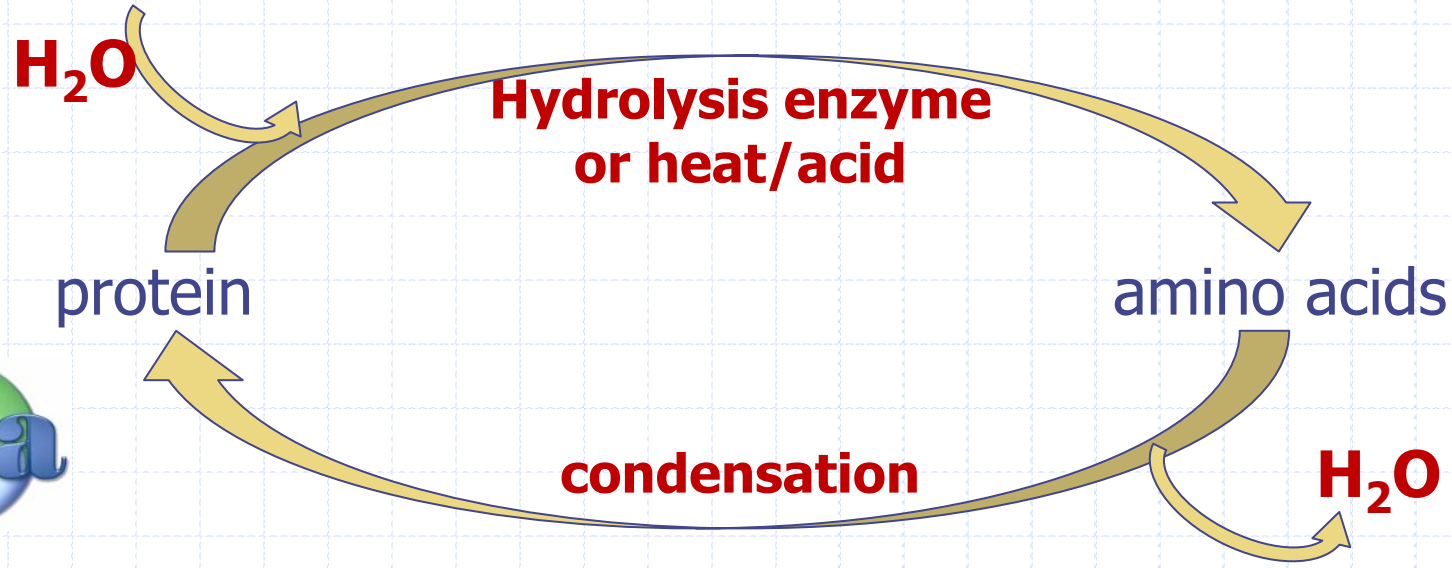


Figure 22.28 Prion protein 'flipping'

B2.5 – Analysis of Proteins

- The analysis of proteins is begun by determining it's amino acid **composition** (*not the primary sequence, the composition.*)
- First you must separate the amino acids from one another by breaking bonds through **hydrolysis** by using acid. This is the opposite of **condensation** used to form peptide bonds
- There are two methods, chromatography and electrophoresis

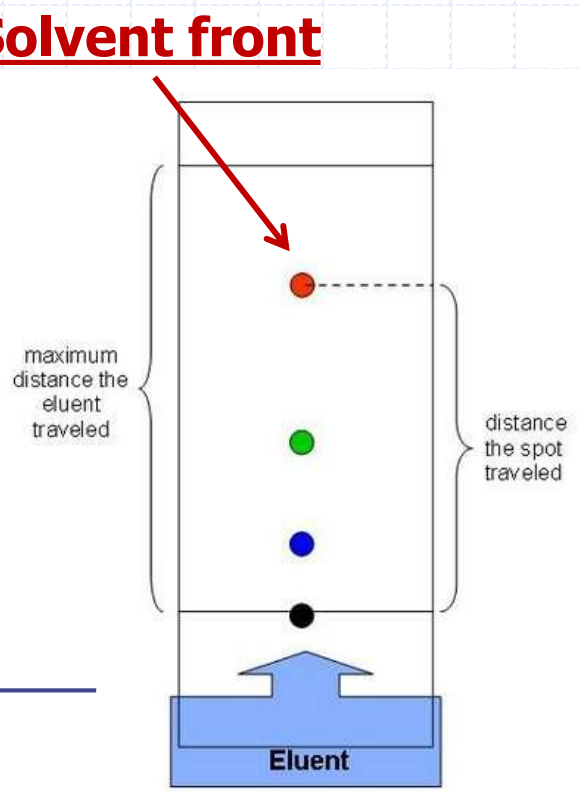


B2.5 – Analysis / Chromatography

- **Chromatography** is a technique used to separate and identify the components of a mixture, particularly when colored
- Amino acids are colorless in solution, but take on color when treated with a **locating reagent**
- Peptide chain must be **hydrolyzed** first as discussed in the previous slide
- The solution is then "spotted" near the bottom of **chromatographic paper** and this position is known and marked as the **origin**
- Paper is then suspended in a chromatographic tank containing a small volume of solvent ensuring that the origin" spot is above the solvent.

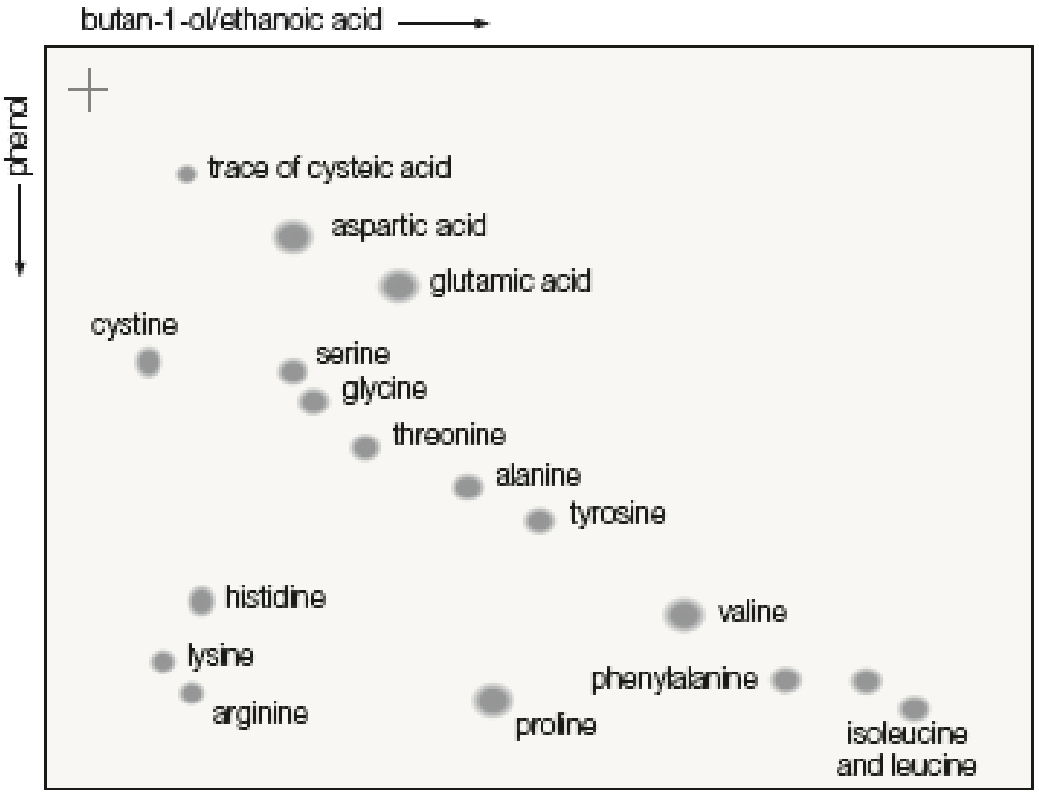
B2.5 – Analysis / Chromatography(2)

- Solvent rises up the paper by **capillary action**, passes through the **origin** and the amino acid distributes between two phases below, and move up the paper at different speeds and are spread according to their solubilities.
  - **Stationary Phase** (water in paper)
  - **Mobile Phase** (the solvent)
- Paper is then removed from the tank and developed by spraying with locating reagent **ninhydrin**, making most of the amino acids a purple color
- Amino acids are then given an $\mathbf{R_f}$ value



B2.5 – Analysis / Chromatography(3)

- A two dimensional (2D) resolution can then be achieved by adding second solvent and rotating the paper 90°

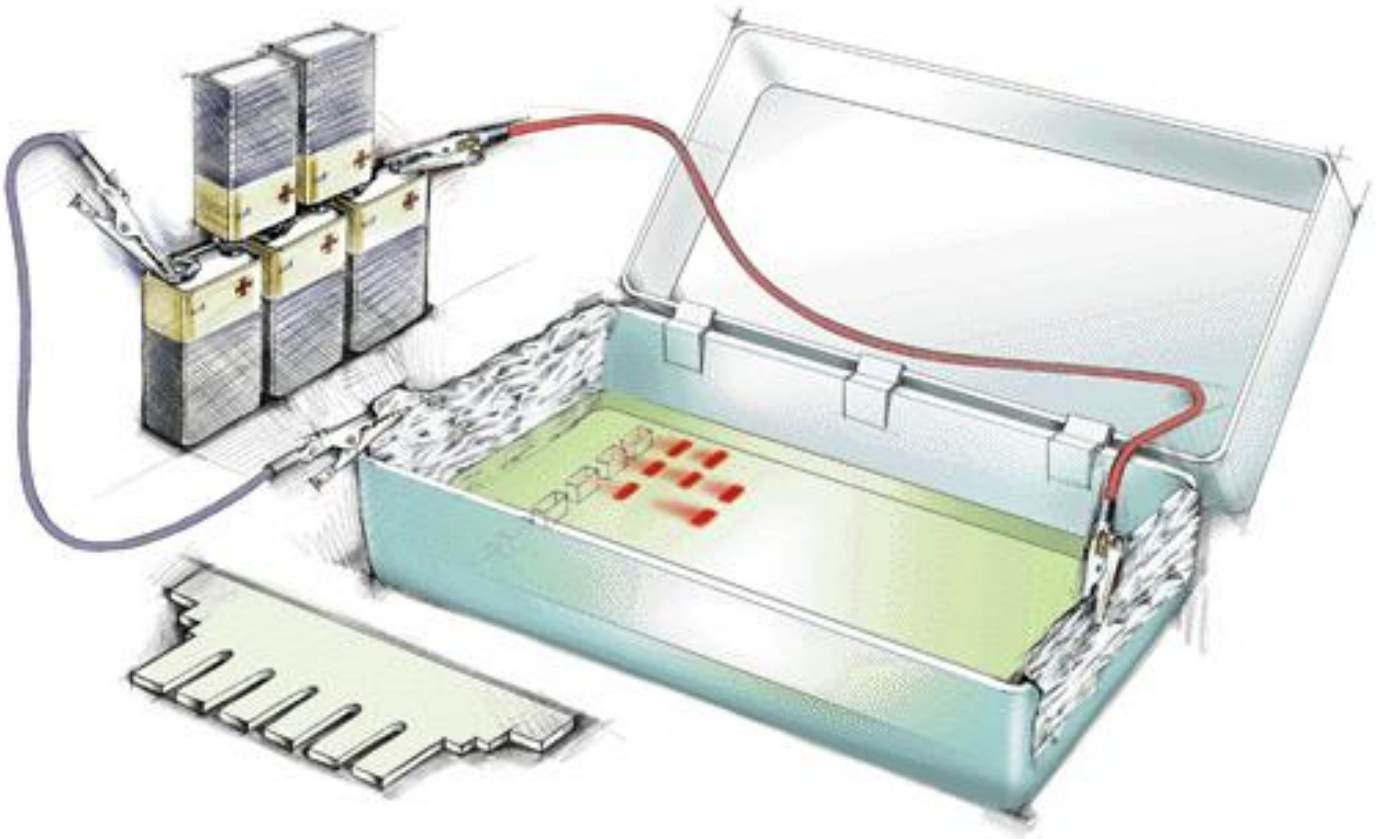


B2.5 – Analysis / Electrophoresis

- Technique used to separate a mixture based on the movement of charged particles in an electric field
- Amino acids carry different charges depending on side chains (R) and their pH so can be separated by these means
- **Gel electrophoresis**, typically made by **polyacrylamide**
- Amino acid mixture is placed in wells and an electric field is applied and AA's move toward electrodes at different rates
- When complete, the amino acids can be detected by stain or made to **fluroesce** (glow) under **UV light**

B2.5 – Analysis / Electrophoresis (2)

- Gel Electrophoresis set up



Learning Check 4

Identify the level of protein structure

1. Electrophoresis 2. Mobile Phase
3. Chromatography 4. Stationary Phase

A. The origin point moves with water

B. Technique used to separate a mixture based on the movement of charged particles in an electric field

C. The origin point moves with solvent

D. Technique used to separate and identify the components of a mixture

Answers 4

Identify the level of protein structure

1. Electrophoresis 2. Stationary Phase

3. Chromatography 4. Mobile Phase

A. (2)The origin point moves with solvent

B. (1)Technique used to separate a mixture based on the movement of charged particles in an electric field

C. (4)The origin point moves with water

D. (3)Technique used to separate and identify the components of a mixture

Learning Check 5

What are the products of the complete hydrolysis of Ala-Ser-Val?

Solution 5

The products of the complete hydrolysis of Ala-Ser-Val are

alanine

serine

valine

Learning Check 6

Tannic acid is used to form a scab on a burn. An egg becomes hard boiled when placed in hot water. What is similar about these two events?

Solution 6

Acid and heat cause a denaturation of protein. They both break bonds in the secondary and tertiary structure of protein.

B2.6 – Functions of Proteins

- Proteins are crucial components for basic life processes
  - Responsible for transport throughout a cell or organism
  - Responsible for maintaining cellular structures
  - Responsible for basic metabolism

B2.6 – Functional Protein Function

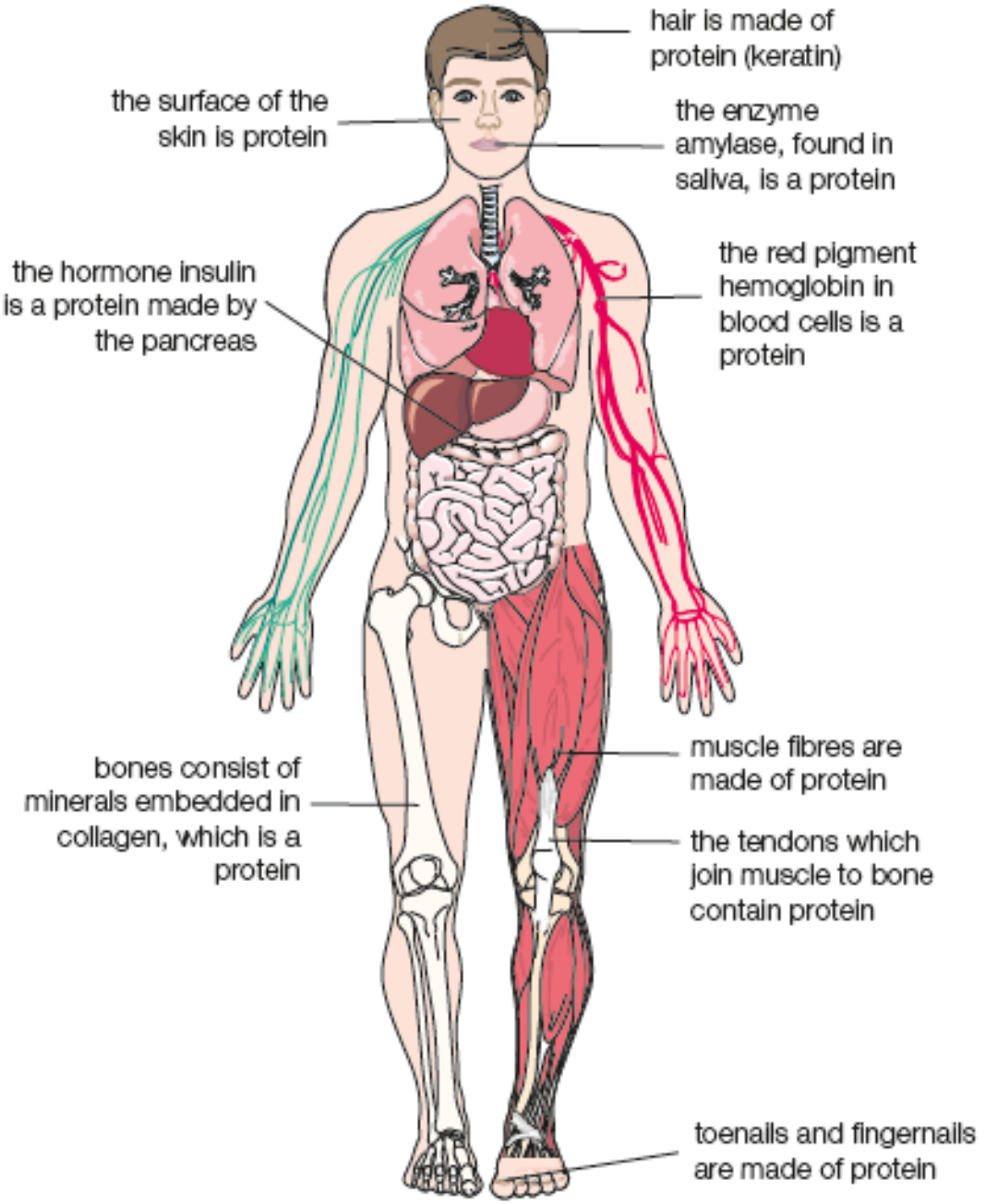
- Functional proteins
  - Transport of oxygen throughout the blood and its storage in muscle cells
  - Completed by hemoglobin and myoglobin

B2.6 – Structural Protein Function

- Structural proteins
  - Elastin is included in the walls of arteries and veins as well as the brochioles (airways) in the lungs.
  - Collagen is the main structural protein of connective tissue in animals and the most abundant protein in mammals. It is the major component of cartilage, ligaments (bone to bone), and tendons (bone to muscle).
  - Muscle is composed of protein, example actin and myosin form the fibers

B2.6 – Protein Function in the Body

- This diagram is meant to aid in connecting your knowledge of chemistry with the use of chemical compounds in the body



B2.6 – Pore Protein Function

- Pore proteins form channels that transport ions, water, and other molecules through the cell membrane.
- Many proteins have enzymatic activity such as catalyzing the digestion of protein, starch and lipids in the gut.
- Digestive enzymes operate outside the cell
- Intracellular enzymes control respiration, photosynthesis (in plants), and DNA repair and replication

B2.6 – Immunoprotein Function

- Immunoproteins (antibodies) are present in blood and identify and neutralize harmful bacteria and viruses.
- Antibodies are produced by a group of white blood cells known as B cells. (HIV infects T cells which regulate the B cells of the immune system.)

B2.6 – Hormone Function

- Hormones are chemical messengers that carry a signal from a cell or group of cells to another via the blood stream.
- All animals and plants produce a variety of hormones.
- In general hormones control the function of their target cells.
- Insulin is a protein-based hormone produced by mammals in the pancreas. Regulates blood glucose levels